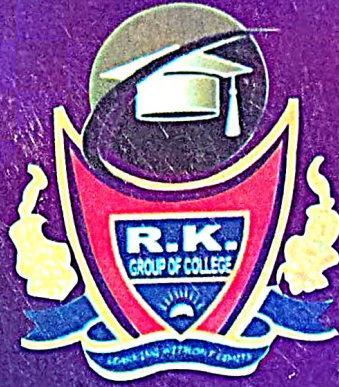
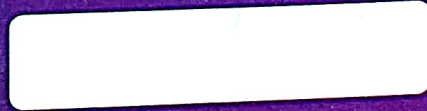




R. K. GROUP OF COLLEGE

BEHIND KALWAR POLICE STATION, KALWAR, JAIPUR (RAJ.)



CERTIFICATE

Name: Radha Yadav

Class: B.Sc. 3rd year

Roll No.:

Exam No.:

Institution R.K. Vigyan P.G. College

This is certified to be the bonafide work of the student in the _____

_____ Laboratory during the academic
year 20 ____ /20 ____

No of practicals certified _____ out of _____ in the
subject of Zoology _____

.....
Examiner's Signature

.....
Teacher in-charge

.....
Principal

Date :

institution Rubber stamp

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Experiment - 1

Introduction → Counting of Red and white Blood Corpuscles in a blood Samples.

Principles → To enumerate RBCs and WBCs draw a blood sample into an RBC or WBC pipette, add the corresponding diluting fluid to mix and dilute the sample, the central 1mm square for RBCs and the four corner squares for WBCs, applying a specific formula for the final count.

Material required → Haemocytometer, Blood Sample, microscope, sterilized needle, Cotton and spirit. etc,

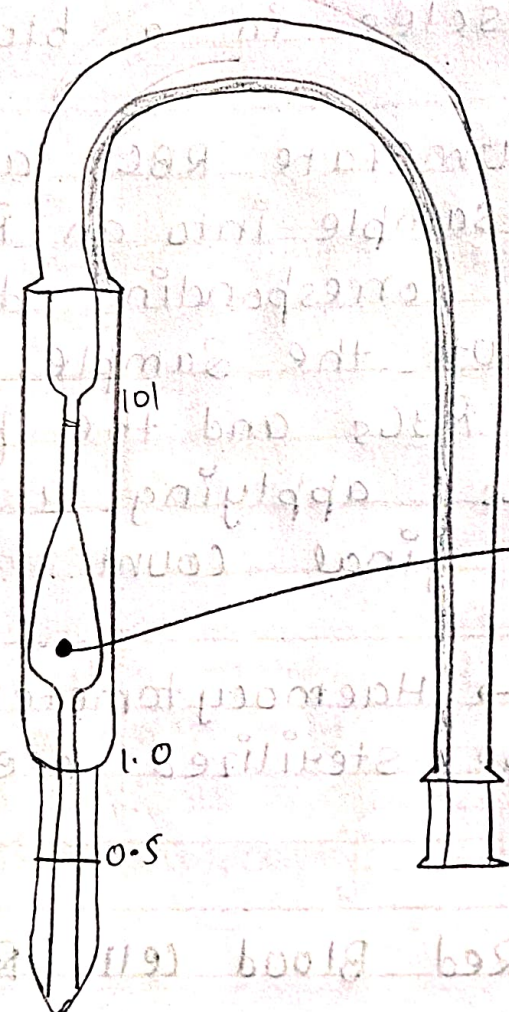
Procedure for Red Blood cell (RBC) →

- (1) **Dilution** → Draw blood into the RBC pipette up to the 0.5 mark, then draw RBC diluting fluid up to the 101 mark.
- (2) **Mixing** → Gently rotate the pipette to mix the blood and diluting fluid thoroughly.
- (3) **Loading** → Place a cover glass on the haemocytometer chamber.

Experiment - 1

Introduction - Counting of RBC and white blood cells in a blood sample

Principle - The sample is placed in a blood pipette and the sample is diluted with a known volume of diluting fluid. The mixture is then counted in a hemacytometer.



Materials required - Hemacytometer, blood sample, microscopical slide, sterile water and spirit, etc.

Procedure for Red Blood Cell (RBC) -

1) Dilution - Draw blood into the RBC pipette up to the 0.2 mark, then 2.0 mark. Diluting fluid up to the 1.0 mark.

2) Mixing - Gently rotate the pipette to mix the blood and diluting fluid thoroughly.

3) Loading - Place a cover glass on the hemacytometer chamber.

(4) Chamber filling → Discard the first drop from the pipette, then touch the tip to the edge of the cover slip, allowing the diluted blood to flow into the chamber by capillary action.

Ensure no air bubbles are present and the chamber is not overfilled.

(5) Settling → Place the haemocytometer on the microscope stage and wait for about 3 minutes to allow the RBCs to settle.

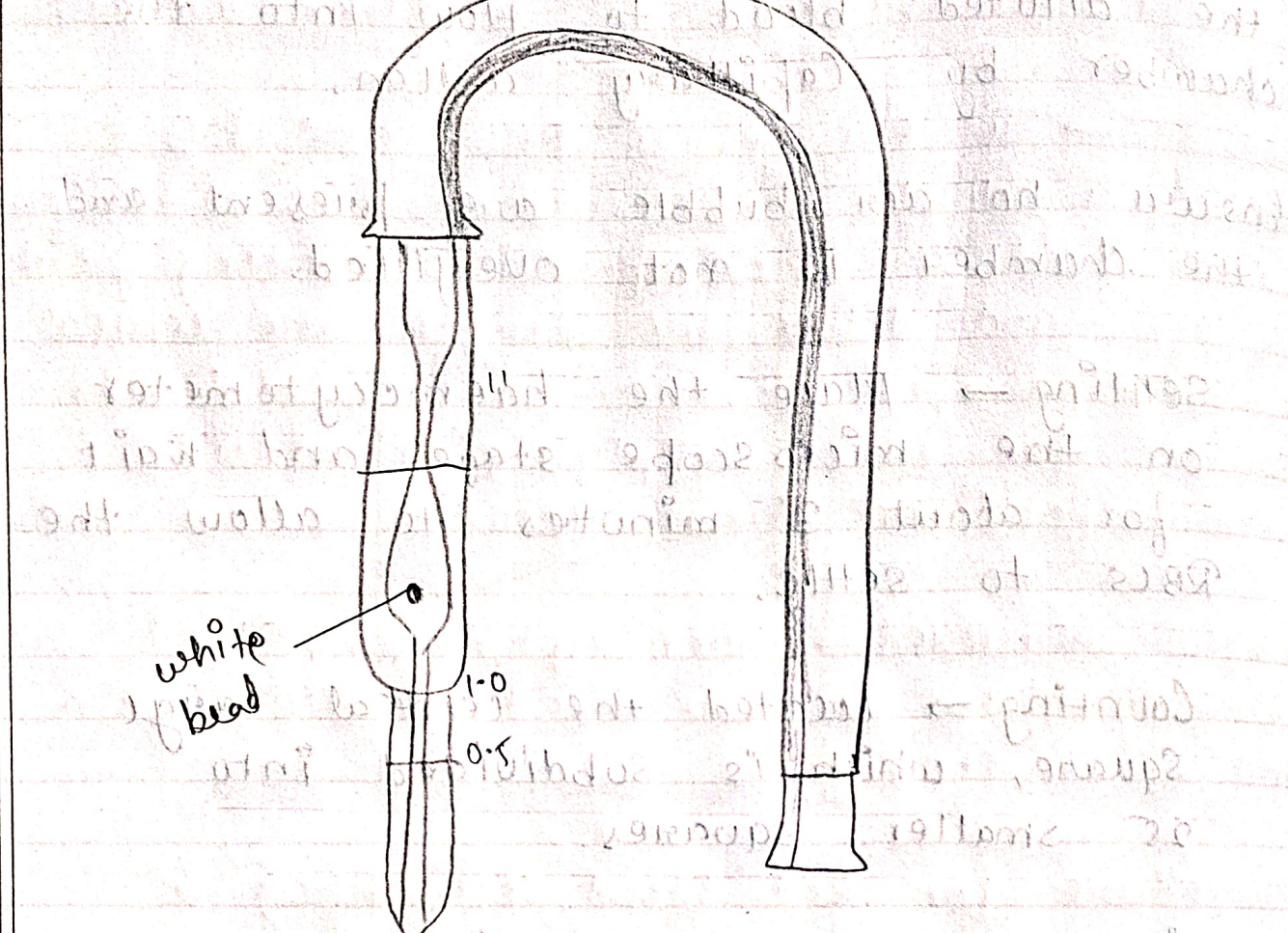
(6) Counting → Located the central large square, which is subdivided into 25 smaller squares.

- Switch to the 40x objective lens.

- Count the RBCs in the four corner squares and one central square of this grid

- Apply the maxian rule → Count cells on the upper and left borders of each square, but not those on the lower and right

Chamber of 1/16" x 1/16" x 1/16" the first drop
 from the pipette, then form the tip
 to the edge of the cover slip, allowing
 the diluted blood to flow into the
 chamber by capillary action.



Counting - The cells are counted in the four corners
 on the middle square stage and then
 for about 2 minutes to allow the
 cells to settle.

Counting - The cells are counted in the four corners
 of the middle square stage and then
 for about 2 minutes to allow the
 cells to settle.

Counting - The cells are counted in the four corners
 of the middle square stage and then
 for about 2 minutes to allow the
 cells to settle.

4) Calculation $\rightarrow$ Multiply the total number of cells counted by the dilution factor (200). Multiply by the depth factor.

• Formula $\rightarrow$ Total RBC Count = $N \times \text{dilution factor} \times 100$ ($N \times 200 \times 50 = N \times 10000$)

Procedure for white blood cell (WBC)

1) Dilution $\rightarrow$ Draw blood into the WBC pipette up to the 0.5 mark, then draw WBC diluting fluid up to the 11 mark.

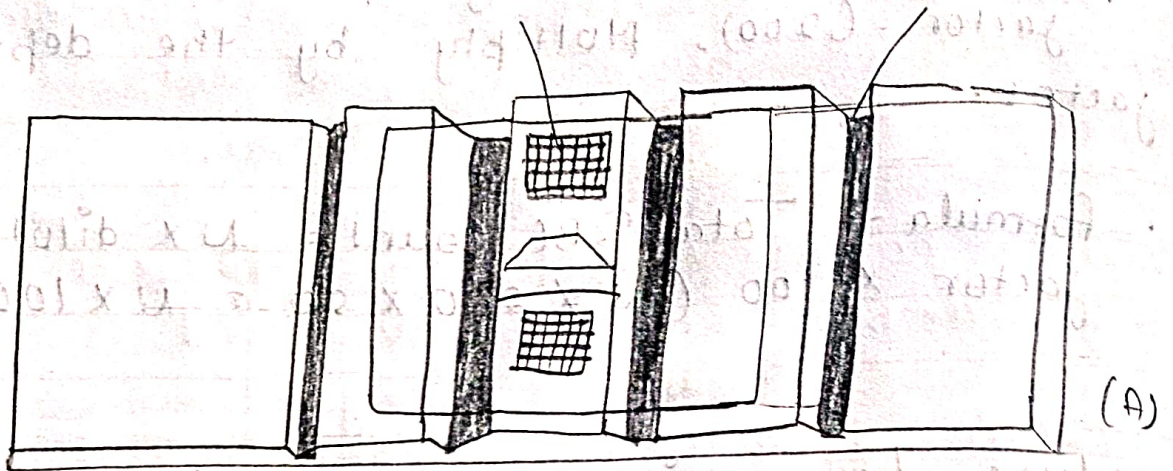
2) Mixing $\rightarrow$ Mix the contents thoroughly by rotating the pipette.

3) Loading $\rightarrow$ Position the cover glass on the haemocytometer and fill the chamber with diluted sample, similar to the RBC procedure.

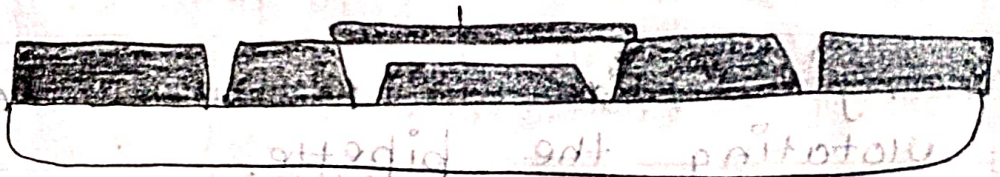
4) Settling $\rightarrow$ Let the chamber rest for a few minutes for WBCs to settle.

Counting Chamber

Coverslip



Coverslip



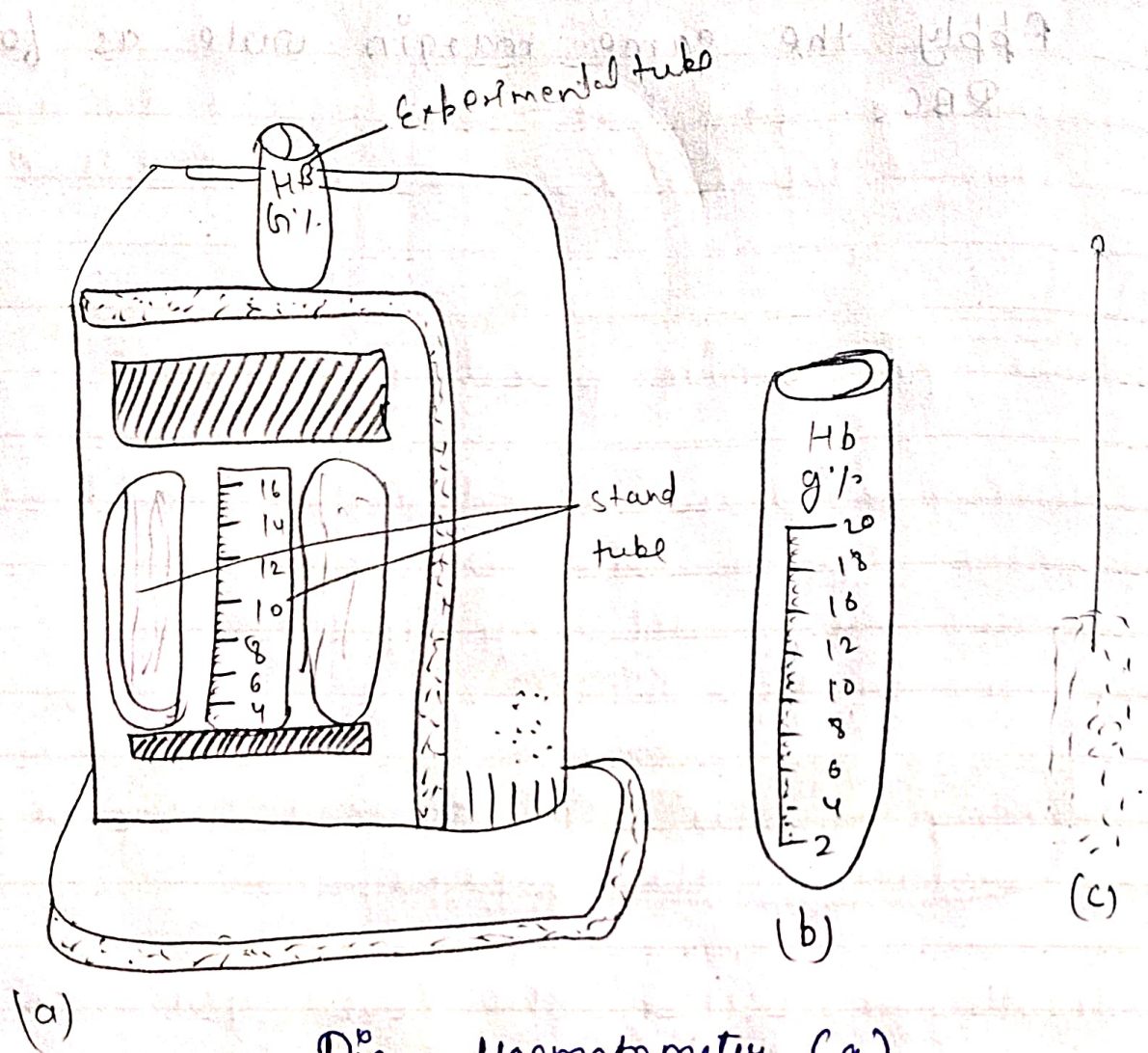
Digital Haematocytometry

the new method will be similar to the RBC haematocrit

settling - let the chamber rest for a few minutes for white to settle

(5) Counting → Locate the four large corner squares on the haemocytometer grid.

Apply the same margin rule as for RBCs.



(a)

Dig → Haemometer (a)

Haemoglobin tube (b)

Experiment → 2

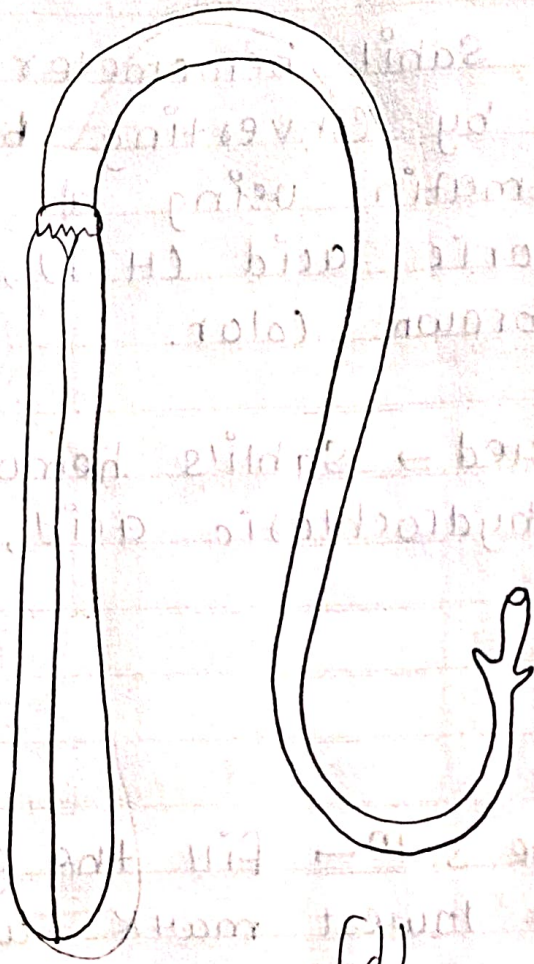
Introduction → Estimation of haemoglobin by Sahli's haemometer.

Principles → The Sahli hemometer estimates haemoglobin by converting blood haemoglobin into acid hematin using a dilute solⁿ of hydrochloric acid (HCl), which produces a brown color.

Material required → Sahli's hemometer set, 0.1N or 1N hydrochloric acid, Distilled water etc.

Procedure →

- (1) Prepare the solⁿ → Fill the graduated tube to the lowest mark with 0.1N HCl
- (2) Collect blood → Sterilize the finger, perform a prick, and use the hemoglobin pipette to draw blood up to the 20 ul mark.
- (3) Mix blood and acid → Transfer the blood into the graduated tube containing the HCl.



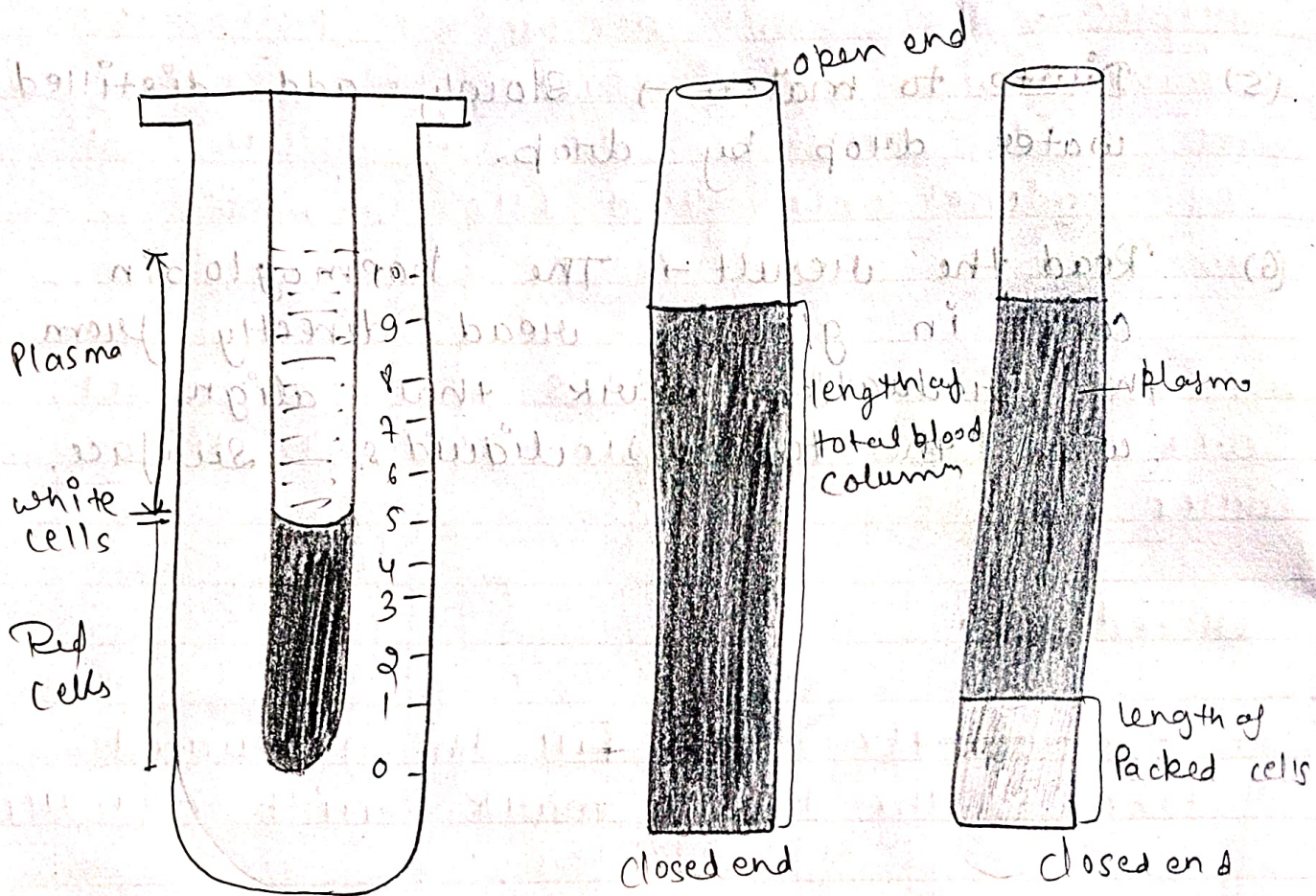
Dig → Pasteur pipette

Mix blood and acid - Transfer the blood into the graduated tube containing the HCl.

- (4) Allow for conversion $\rightarrow$ Let the mixture stand for 5 to 15 minutes to allow the hemoglobin to convert to acid hematin.
- (5) Dilute to match $\rightarrow$ Slowly add distilled water drop by drop.
- (6) Read the result $\rightarrow$ The haemoglobin concⁿ in g/dl is read directly from the graduation mark that aligns with the top of liquid's surface.

DATE NAME

Teacher's Signature.....



Dia → Hemocrit tube

Experiment - 3

Introduction → Haematocrit Value or Packed cell Volume, PCV.

Principles → The packed cell volume (PCV), is estimated by dividing the Volume of red blood cell (RBCs) by the total volume of blood and multiplying by 100.

step by step manual management →

(1) collect a blood sample → A small blood sample is drawn into a capillary tube. The tube is then sealed at one end.

(2) Centrifuge the sample → The sealed tube is placed in a microhematocrit centrifuge and spun at a high speed for several minutes.

(3) Take the measurement → using a special reader card or a ruler, measure the height of the RBC layer and the total height of the blood column.

(4) Calculate the PCV →

$$PCV = \frac{\text{Height of RBCs}}{\text{Total height of blood}} \times 100$$

Normal Ranges

- Men $\rightarrow$ 40 - 54%.
- Women $\rightarrow$ 36 - 48%.
- children $\rightarrow$ 32 - 44%.
- Newborn $\rightarrow$ 44 - 64%.

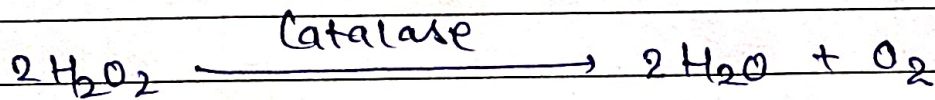
Experiment → 4

Introduction → Demonstration of Enzyme Activity (Catalase) in Liver

Principle → Liver is an ideal source for this experiment because it has one of the highest concentration of Catalase in the body.

The Catalase reaction →

Catalase breaks down hydrogen peroxide (H_2O_2) into water (H_2O) and oxygen gas (O_2)



material required → Fresh Liver, Ruler, Test tube, small beakers.

Basic set up →

- 1) Prepare the liver extract → mince or blend a small amount of liver with an equal volume of water to create a uniform extract.

- (2) Add hydrogen peroxide → Pour a small amount of hydrogen peroxide into a test tube.
- (3) Initiate the reaction → Add a few drops of the liver extract to the hydrogen peroxide.
- (4) Result → The rapid and vigorous bubbling indicates the production of oxygen gas, confirming the presence of active catalase.

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Experiment - 5

Introduction → Study the effect of pH and temperature on salivary ~~glamylase~~ amylase activity.

Principle → High temperature denature the enzyme, while low temperatures render it inactive, and extreme pH levels cause denaturation and a loss of function.

Effect of temperature →

- **Optimal temp.** → The enzyme functions most effectively at 37°C , which is the body's temp.
- **Below optimal** → At temp. lower than 37°C the reaction rate slows down.
- **Above optimal** → As temp. rises above 37°C , the enzyme's structure is altered.

Procedure →

1. Prepare starch solutions and incubate them at different temp. or with different buffer solutions.

2. Add saliva to each solution.
3. Take samples from each tube at regular intervals and test for the presence of starch using iodine.
4. The tube where the blue-black color disappears first has the fastest enzymes activity.

Experiment - 6

Introduction → Detection of proteins in the animal tissue / food samples.

Principle → A range of analytical techniques are used, varying from simple qualitative tests to complex, highly sensitive instrumental methods.

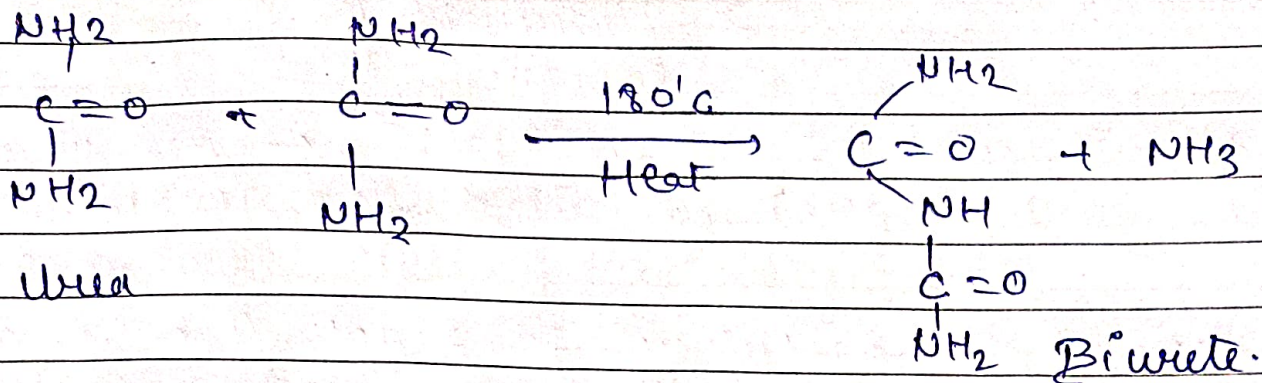
Detection of proteins →

(1) **Biuret test** →

material → sprits, trumps.

- It indicates the presence of peptide linkage in proteins.
- Test is use for detection of Protein/ peptides.

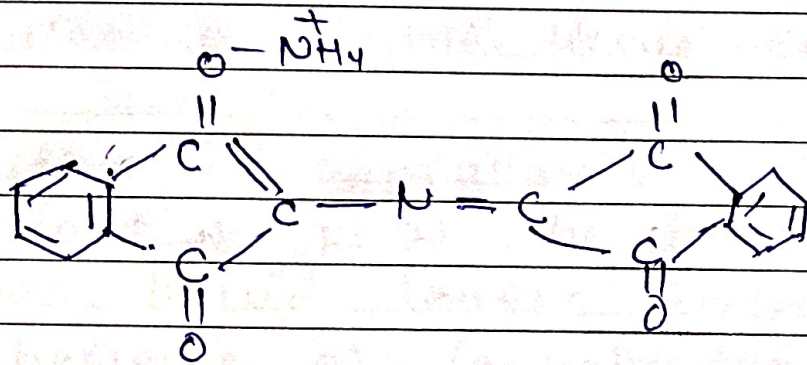
Principle → Cupric ions of Copper sulphate solutions in alkaline medium form coordinate complex with at least two nitrogen of the peptide bonds to form purple colored complex.



(2) Ninhydrin test →

- This is given by all compounds having free α -amino groups.
- Indication/ Purple complex.

Principle → Ninhydrine + α -amino acid →
 hydrindantin + aldehyde + CO_2 + NH_3 Hydrindantin + NH_3 + Ninhydrine → violet colored.



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Experiment - 7

Introduction → Detection of carbohydrates in the animal tissue / food samples.

Principles → Both qualitative and quantitative methods can be used to detect and analyze carbohydrates in animal tissue food samples.

1) Molisch's test →

This test uses α -naphthol and concⁿ sulphuric acid to detect nearly all carbohydrates.

Procedure → Add a few drops of molisch's reagent to the tissue extract.

Result → The appearance of a purple or violet ring at the junction of the two liquid layers indicate the presence of carbohydrates.

2) Benedict's test →

This test identifies the presence of reducing sugars like glucose and fructose.

Procedure → Mix benedict's reagent with the tissue extract and heat the mixture in a boiling water bath.

Result → A Colored precipitate will form, changing from green to yellow to brick-red depending on the concentration of reducing sugar present.

Experiment - B

Introduction → Detection of lipids in the animal tissue / food samples.

Tests for lipids → Medical tests like a lipid profile to measure blood cholesterol and triglycerides, and qualitative lab tests like the emulsion.

(1) Emulsion test →

(1) The tissue sample is first mixed with ethanol.

(2) The ethanol solution is poured into water.

(3) A +ve result is the formation of a milky white emulsion.

(2) Solubility test →

Requirements →

Sample solution.

Solvents (HCl, NaOH, Water)

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material required →

test tube, test tube stand, Dropper.

Experiment - 9

Introduction → Demonstration of separation of amino acids from a mixture using paper chromatography / TLC.

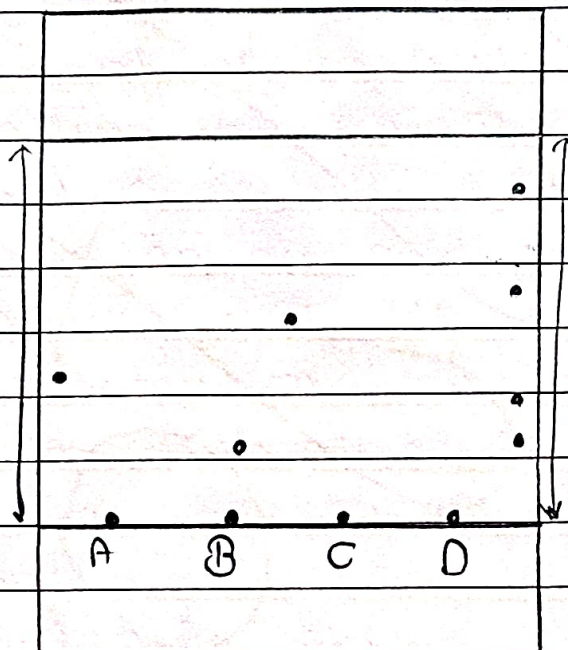
Practical material → chamber of paper chromatography, Watman no. 1, filter paper, butanol.

Procedure → Paper chromatography separates a mixture of amino acids by their different affinities for a stationary phase (paper) and a mobile phase (solvent). A spot of the amino acid mixture and known amino acids is placed on a pencil line on chromatography paper. The paper is then on ~~chromatograph~~ cylinder and placed in a beaker with a mobile phase, ensuring the spot is above the solvent level. As the solvent moves up the paper wth capillary action, it carries the amino acids with it, separating them based on how far they travel.

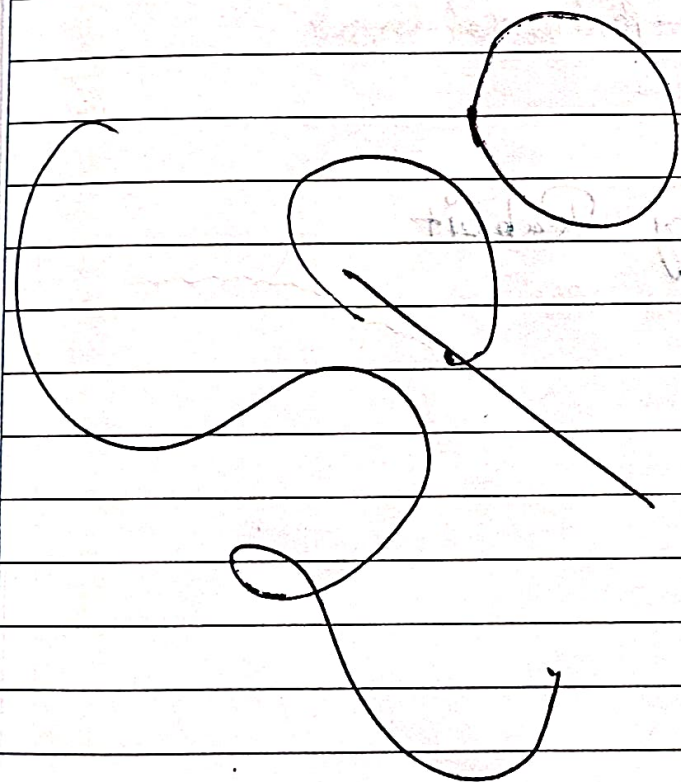
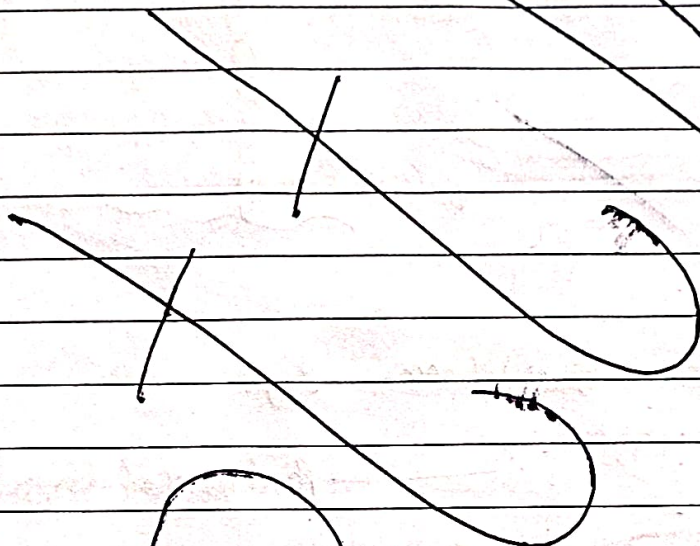
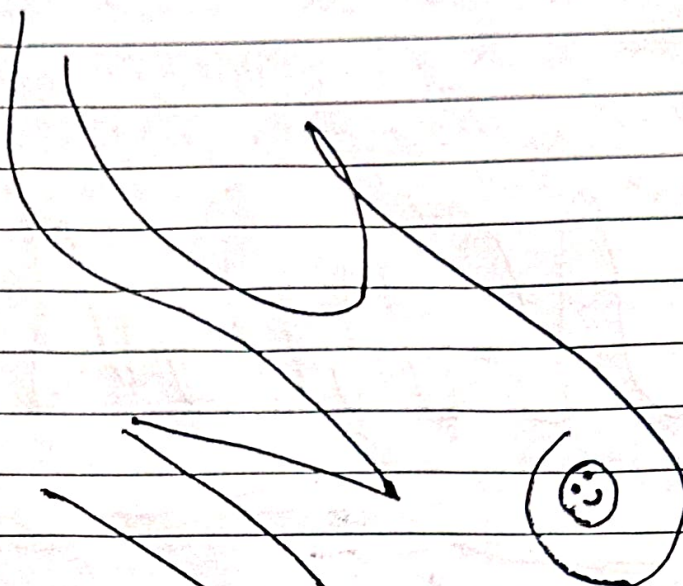
The components are then visualized by spraying with a reagent. Like

min hydriin, which turns amino acids purple, and their identities are determined by comparing their retention factor (R_f) value to those of the known amino acids.

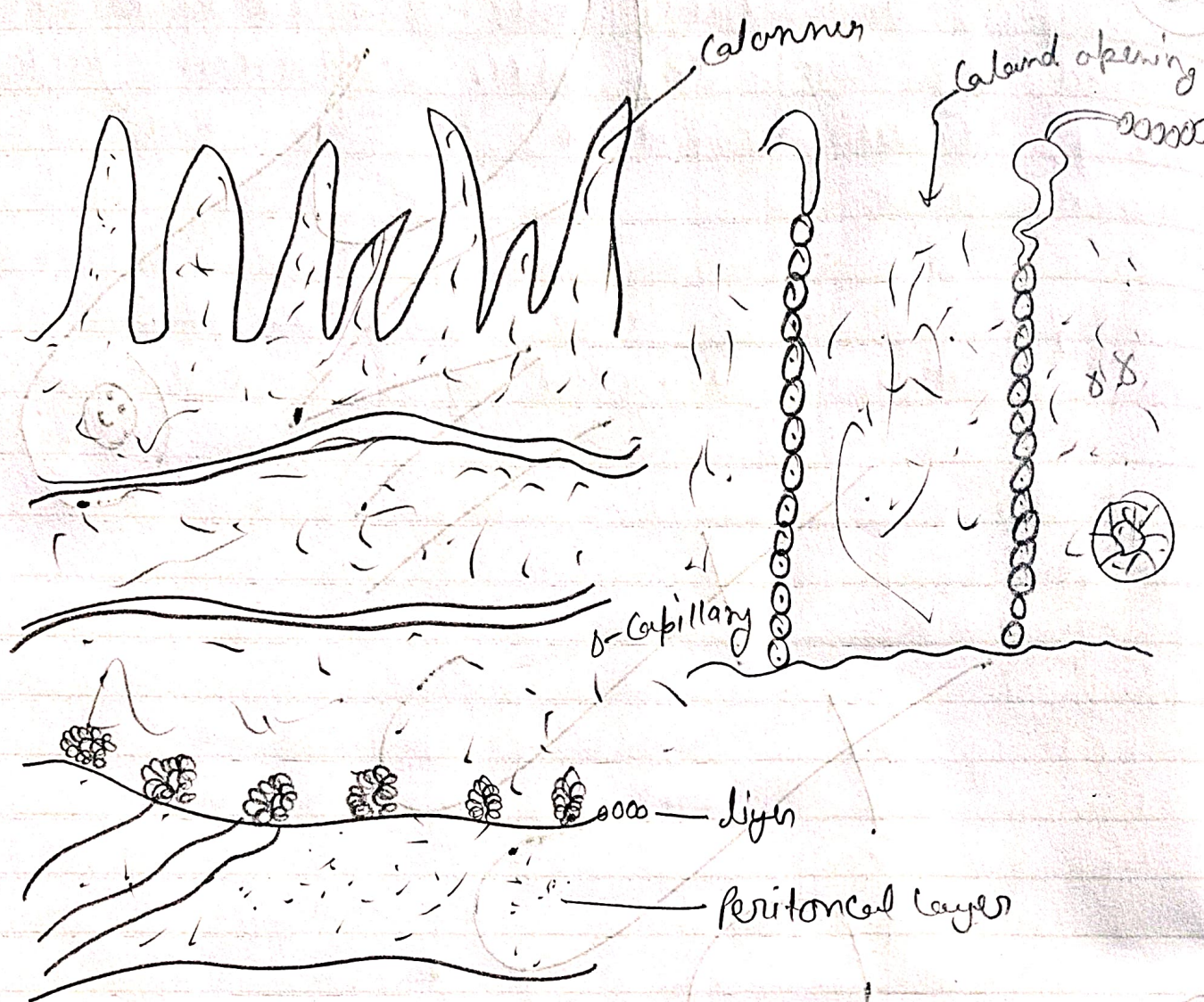
$$R_f = \frac{\text{Distance traveled by the spot}}{\text{Distance traveled by the solvent front.}}$$



paper chromatography



THE END IS ALWAYS A SURPRISE

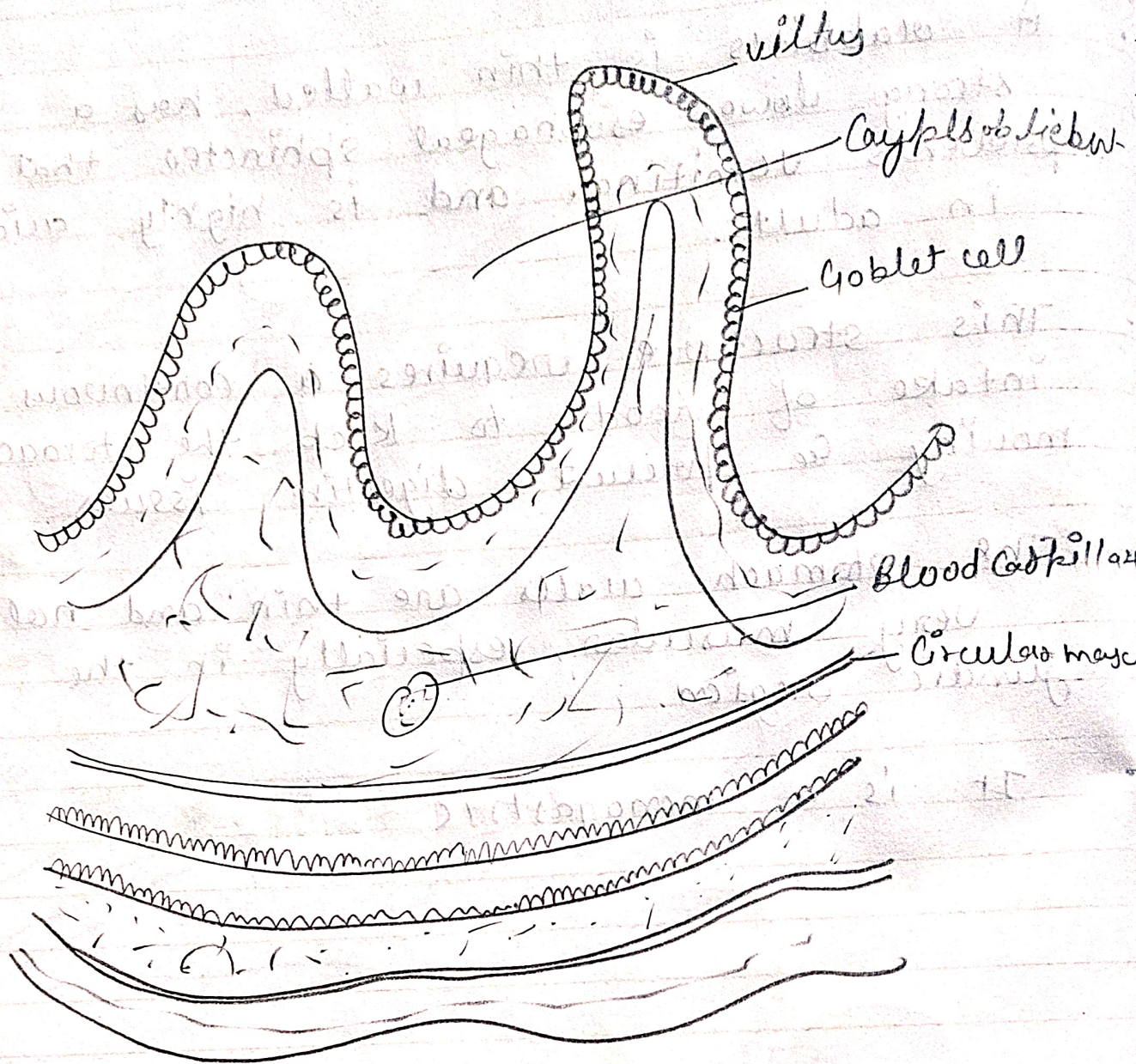


Dig & stomach of Rabbit

Stomach

- A rabbit's is thin walled, has a strong lower esophageal sphincter that prevents vomiting, and is highly acidic in adults.
- This structure requires a continuous intake of food to keep the stomach moving to prevent digestive issues.
- The stomach walls are thin and not very muscular, especially in the fundic region.
- It is a monogastric.

Stomach is not very acidic

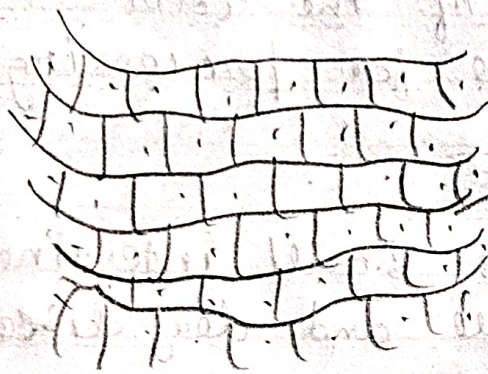


Dig → Intestine of Rabbits.

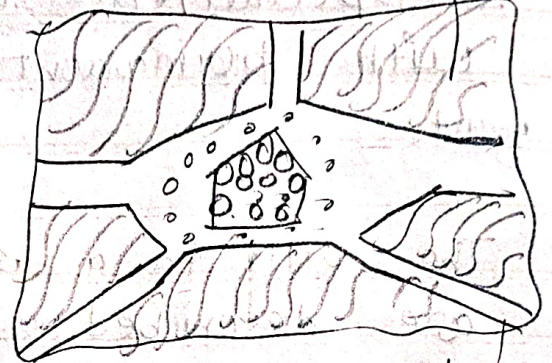
Intestine

- A Hindgut fermenter, a large and well differentiated large intestine, and a unique colon that efficiently separates digestible and indigestible food material.
- A specialized section of the colon with permanent folds for processing food.
- The villi in a rabbits small intestine are described as tall and leaf like.

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Hepatic
cells



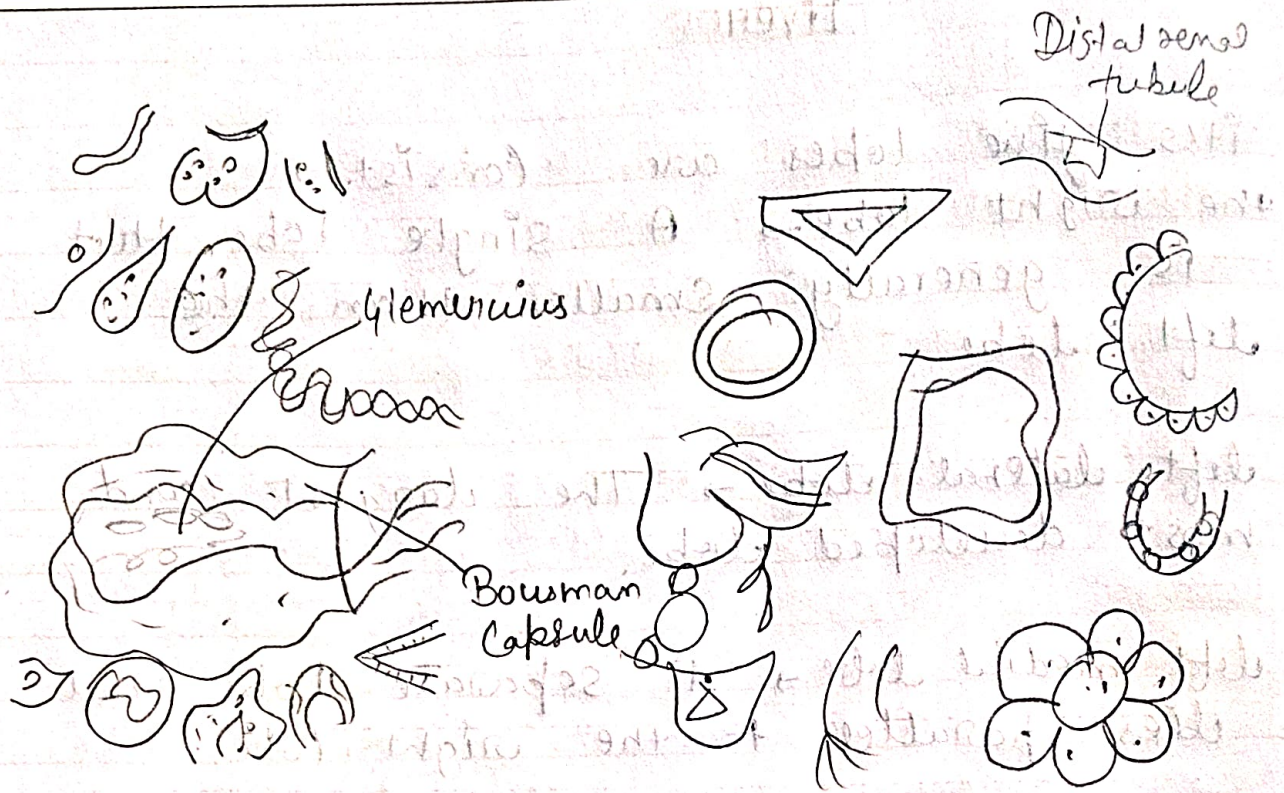
Hepatic
lobule

Dig → liver of rabbit

Liver

- Its five lobes are consist. →
 - (1) the right lobe → A single lobe that is generally smaller than the left lobe.
 - (2) left lateral lobe → The largest and most developed lobe.
 - (3) left medial lobe → A separate lobe that lies parallel to the right lobe.
 - (4) Quadrate lobe → The smallest lobe, located between the left and right lobes.
 - (5) Caudate lobe → A separate lobe with a narrow attachment to the liver's hilar region.

liver is made up of 5 lobes

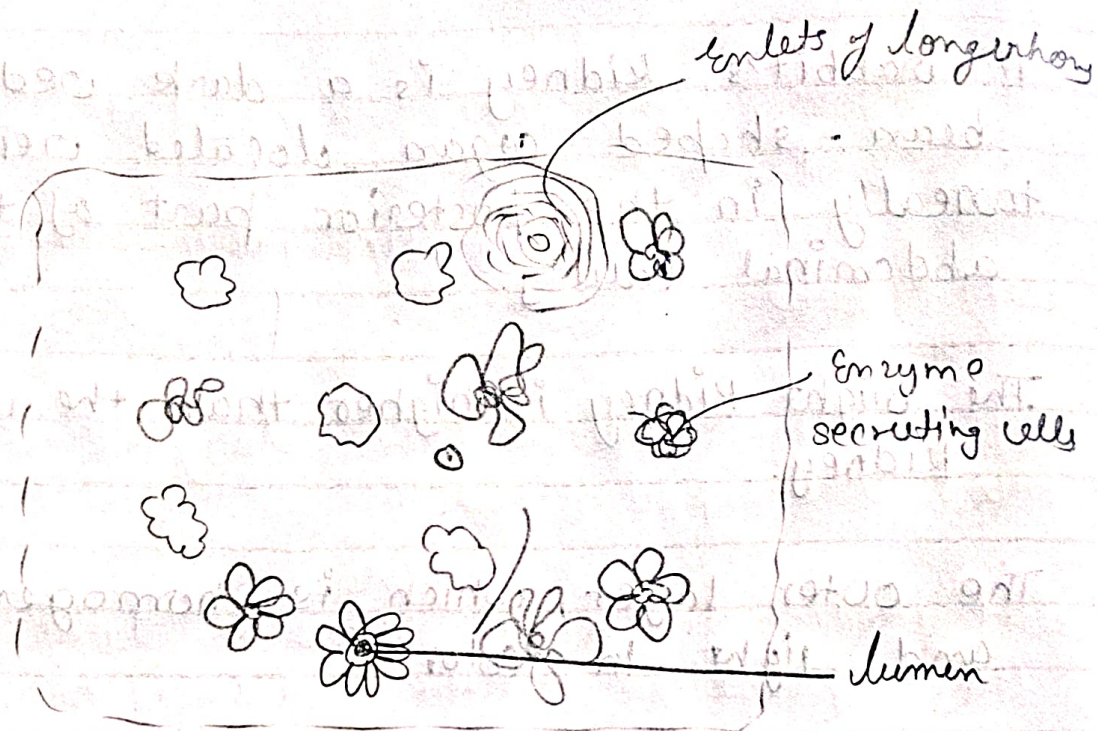


Dr. H. Kidney of rabbit

Kidney

- A rabbit's kidney is a dark red, bean-shaped organ located retroperitoneally in the posterior part of the abdominal cavity.
- The right kidney is higher than the left kidney.
- The outer layer, which is homogenous and light in color.
- The inner layer, which is darker in color and composed of several conical lobes called pyramids.

pancreas



Dig's Pancreas

Pancreas

- The head, body and tail located in the mesentery between the U shaped duodenum.
- It produces and secretes pancreatic juice containing digestive enzymes and bicarbonate ions.
- These include trypsin, amylase and lipase.

but lipid? - yes

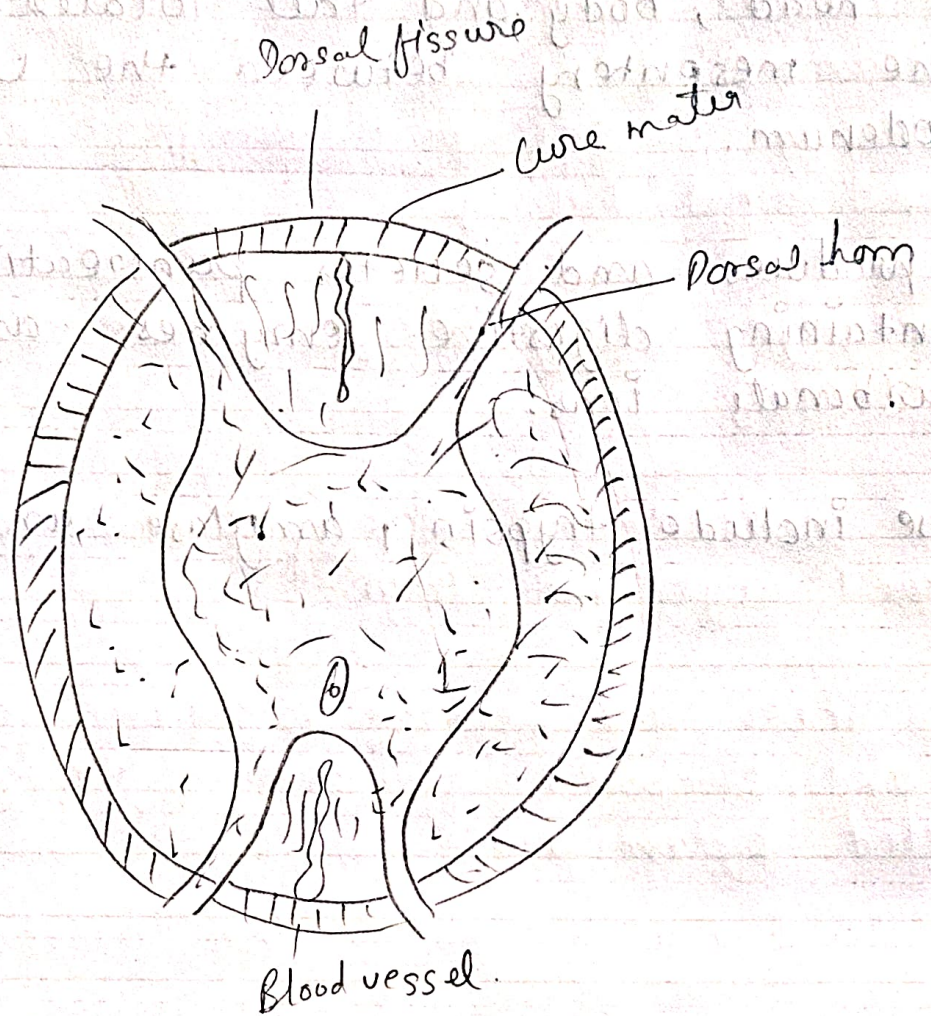
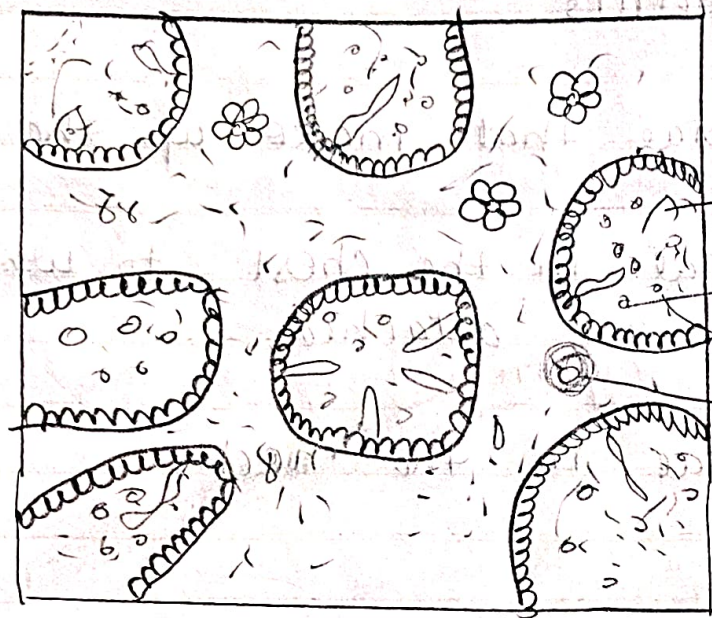


Fig -> Spinal cord

Spinal Cord

- four vertebrae fused together to form the sacrum, which is attached to the pelvis.
- 15 Vertebrae that make up the tail.
- 12 vertebrae in the chest, to which the ribs are attached.
- 7 vertebrae in the neck.

Spinal Cord

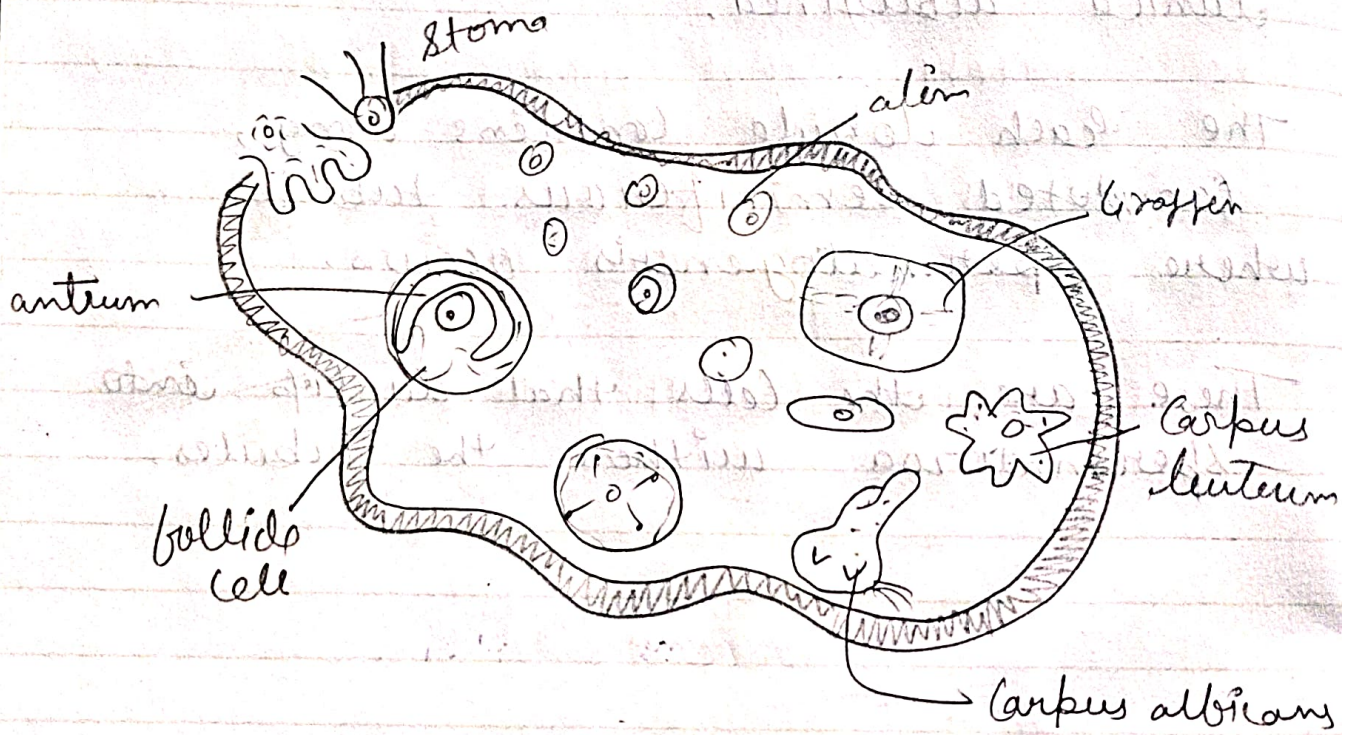


Dig → testis

testis

- Testis are pink, ovoid bodies located in the scrotum, covered by a tough fibrous capsule called the tunica albuginea.
- The each lobule contains long, convoluted seminiferous tubules where spermatogenesis occurs.
- These are the cells that develop into spermatozoa within the tubules.

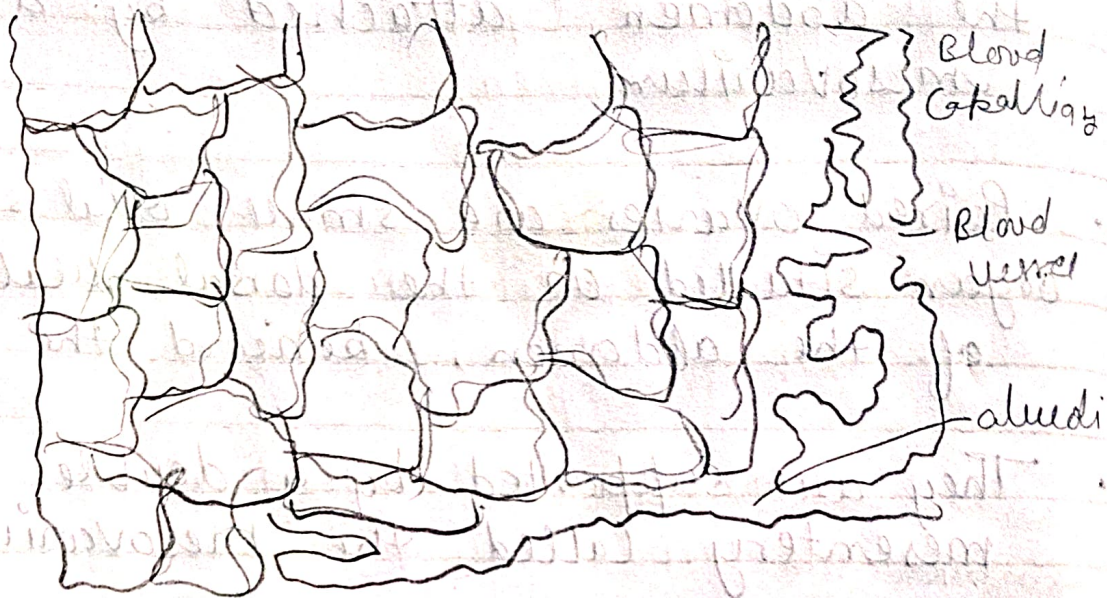
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Pig → Ovary

Ovary

- An oval-shaped, whitish body located on the dorsal wall of the abdomen, attached by a mesovarium.
- Paired ovaries are small, oval shaped organ situated on the dorsal wall of the abdomen, behind the kidneys.
- They are supported by a double fold of mesentery called the mesovarium.
- The outermost layer is the germinal epithelium, made of simple cuboidal cells.



Dig - lungs

Lung

- The pair of lungs is situated within the thoracic cavity, on either side of the heart.
- The lungs are enclosed in a membrane called the pleural cavity.
- They are soft & spongy.

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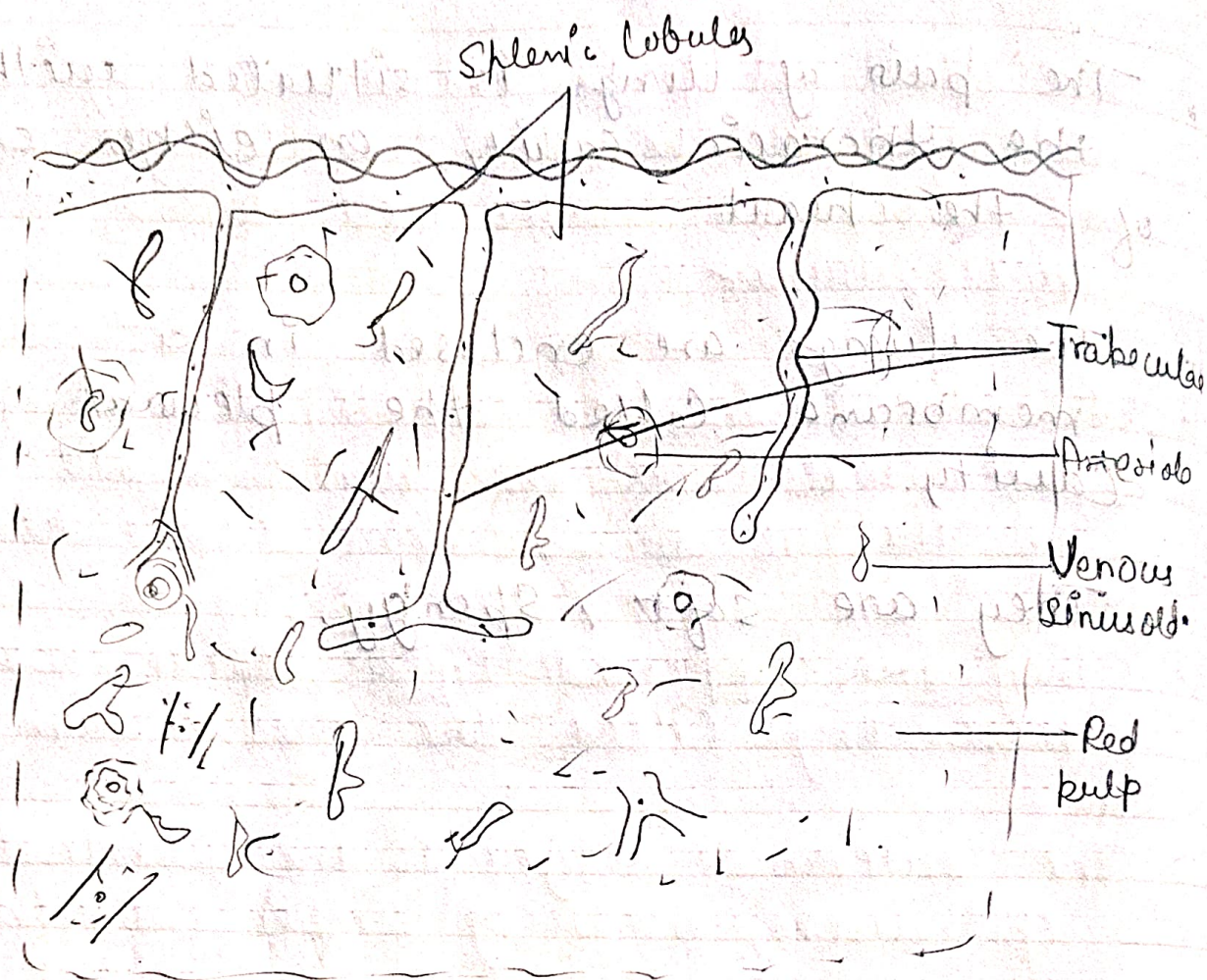


Fig → T.S. of Spleen

Spleen

- It is a soft, friable, dark purple bean-shaped organ.
- A healthy adult spleen is typically 7 to 14 cm long & weighs 75 to 120 grams.
- A fibrous outer layer that protects the spleen and allows for expansion.
- Contains lymphocytes that are crucial for immune responses.

ब्रह्म लक्षणो १० २.३५ - ३.३५

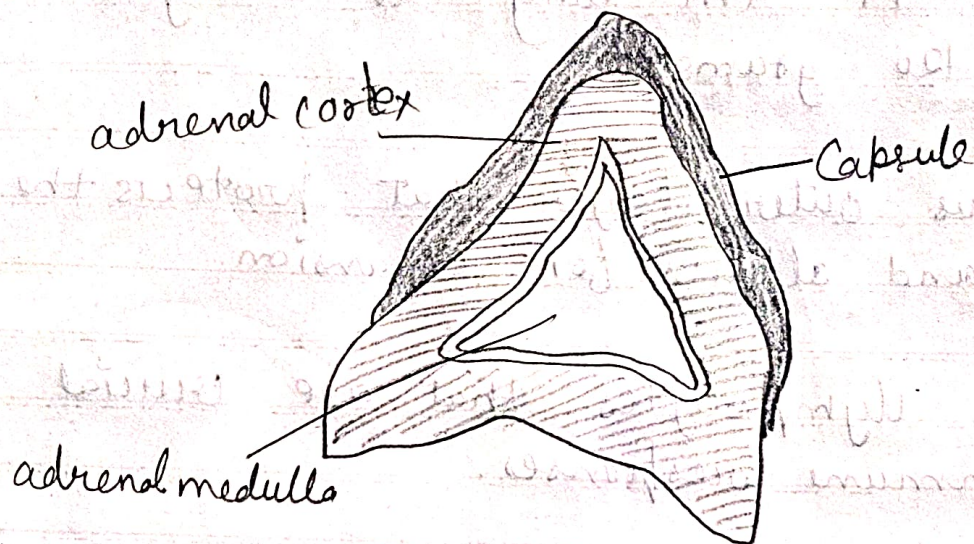


Fig. → T.S of adrenal gland

Adrenal Gland

- situated superior to each kidney.
- It is a triangular, though the right gland is often described as pyramidal and the left is semilunar.
- Its color is yellowish.
- In this a two layers are consists → adrenal cortex & adrenal medulla

biomergent to 217 - gift

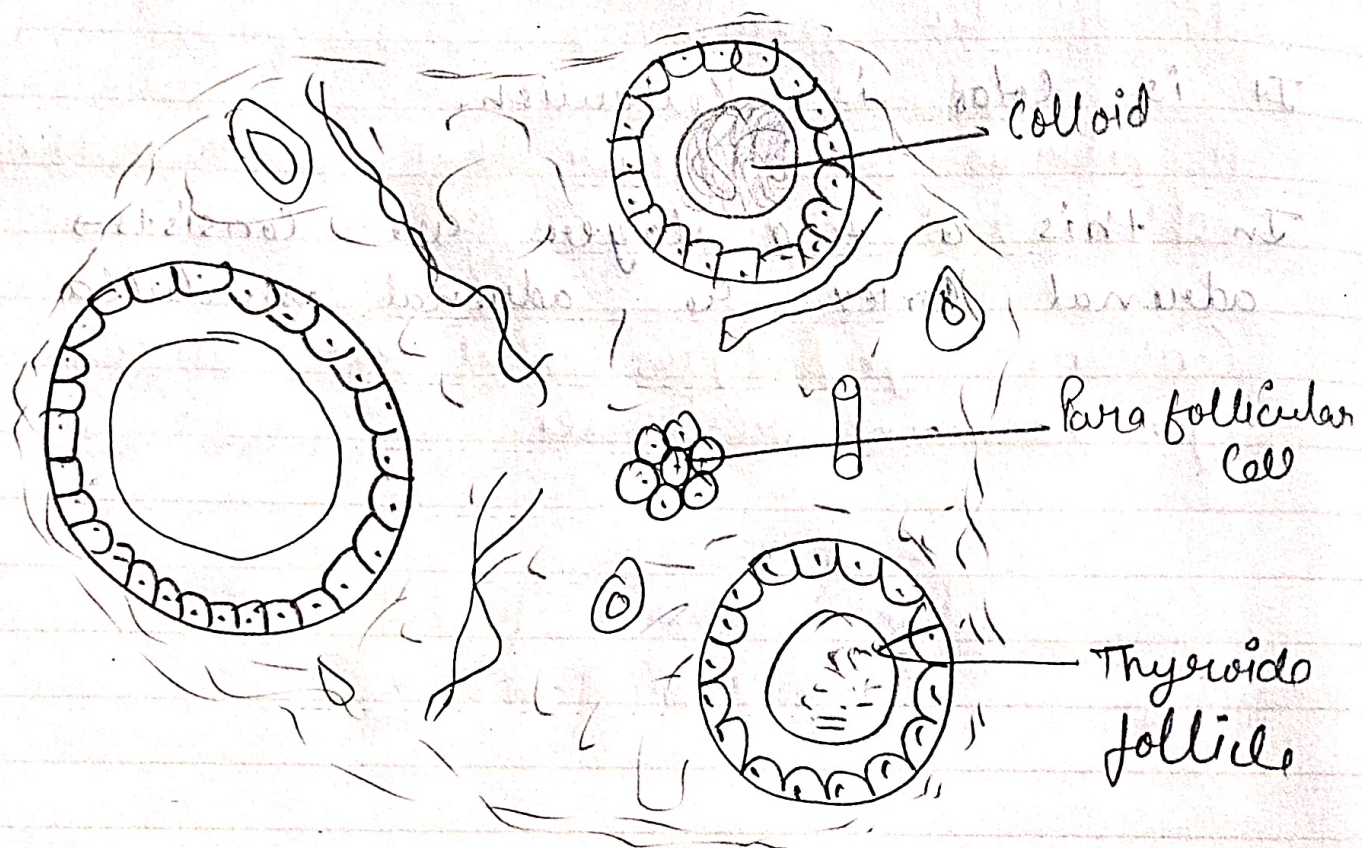


Fig - T.s. of thyroid

Thyroid

- It is a butterfly shape and location at the front of the neck, producing hormones that control metabolism.
- It is located in the front of the neck, just below the Adam's apple.
- That regulated the body's metabolism, influencing functions like heart rate, digestion, & body temperature.

biomaterials pg. 27-31

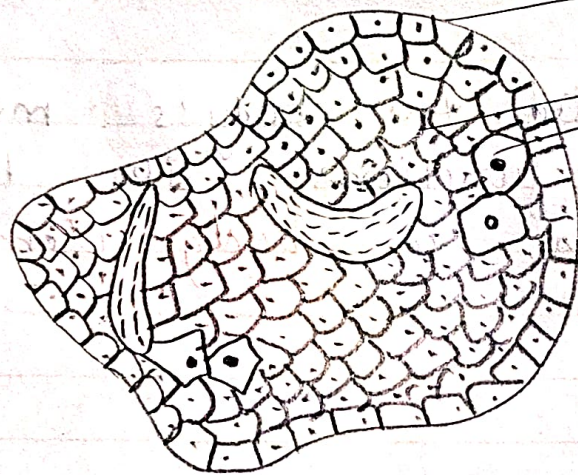
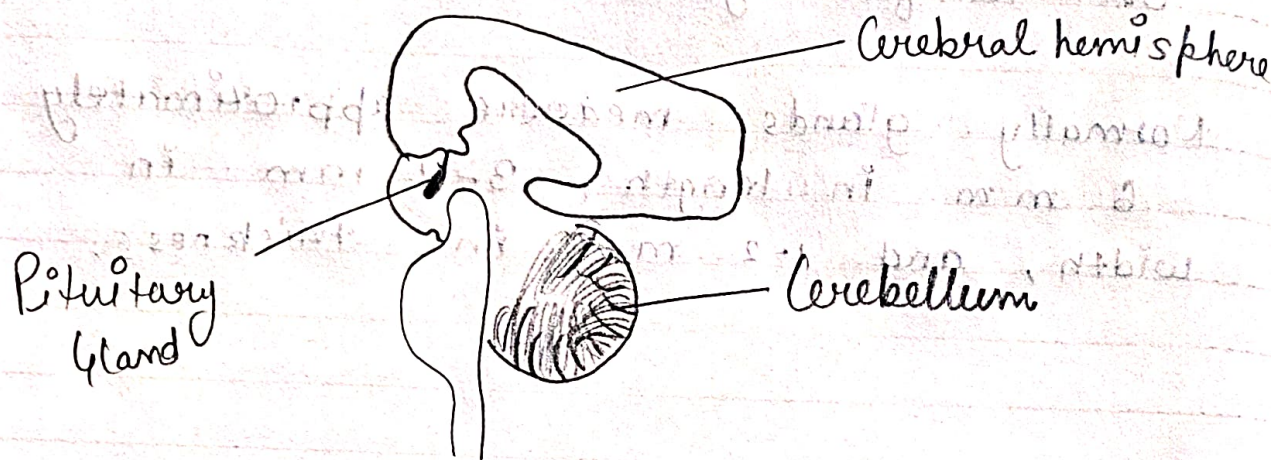


Fig → T.S. of Parathyroid

Parathyroid

- It is a small, pea-sized, ovoid endocrine gland. located behind the thyroid, which typically consist of four gland.
- It is a yellowish-brown, ovoid or lentiform gland.
- Normally glands measure approximately 6 mm in length, 3-4 mm in width, and 1-2 mm in thickness.

basophilic granular gland



Dig - pituitary gland

Pituitary

- Small, pea-sized gland located in a bony depression at the base of the skull, just behind the bridge of the nose.
- Anterior lobe secretes the majority of pituitary hormones.
- Posterior lobe stores and releases hormones produced by the hypothalamus.
- It is the ^{bony} ~~bony~~ cavity is called the sella turcica.